would ever be expected to occur in the future in a natural aquatic ecosystem. In this study, microcosm and mesocosm nanotoxicology studies in the marine environment were conducted focusing on realistic exposure conditions in the ng/L range. An aqueous suspension of branched poly(ethylenimine) capped silver nanoparticles (AgNPs) of 60 nm size was used for this AgNPs exposure study. Preliminary analysis revealed that, in the presence of silver nanoparticles, there was a decrease in enzymatic antioxidant activity, photosynthetic activity and chlorophyll concentration and an increase in intracellular ROS formation and lipids peroxidation in planktonic communities which indicate that the organisms possibly underwent oxidative stress.

MO289

Size of agglomerates like a major contribution to silver nanoparticles aquatic toxicity

J. Oprsal, University of Pardubice / Institute of Environmental and Chemical Engineering; M. Pouzar, University of Pardubice / Environmental and Chemical Engineering; P. Knotek, University of Pardubice; L. Blaha, Masaryk University, Faculty of Science / Faculty of Science RECETOX; P. Plodikova, Research Institute of Organic Synthesis

One of the general problems in aquatic toxicity testing of nanoparticles is their fast agglomeration and subsequent sedimentation, which limits the use of "standardized" protocols in the assessment of particle toxicity. The present work suggests an original approach enabling to control the maximal average size of nanoparticle agglomerates during the aquatic toxicity test. The modification is based on the variable frequency of exposure media exchange, i.e. frequent exchanges of the tested colloid depending on the nanoparticle concentration levels. The method was applied for the short-term toxicity test with the embryo and sac-fry stages of common carp *(Cyprinus carpio)*. Toxicity of silver nanoparticles (AgNPs) with initial average hydrodynamic diameter (D_H) of 40 nm was tested at four concentrations (5; 10; 25 and 50 mM). Based on the chemistry and behavior of AgNPs in the culture medium, two levels of maximal average sizes of agglomerates (200 and 400 nm) were selected, and exchanges of media were employed to control for formation of these types of agglomerates. The comparisons with the semi-static fish embryotoxicity test, where the formation of agglomerates had not been controlled, indicate that different sizes of agglomerates influence different stages of embryo development. Interestingly, larger agglomerates rather than original nanoparticle colloids were responsible for the most severe effects, which could be mechanistically explained by other observations during the study. Details on the novel testing approach and its advantages and limitations will be discussed.

MO290

Effect of silver nanowires on embryonic development of Daphnia galeata

C. Rongxue, Konkuk University; Y. An, Konkuk University / Department of Environmental Science

Silver nanomaterials were widely used in various nanoproducts. This study investigated the effect of silver nanowires (AgNWs) using water flea *Daphnia geleata* embryo. Two types of AgNWs tested were 10 um- and 20 um- AgNWs having different length. Stage two embryos were harvested from adult *Daphnia geleata* female which completed third reproductions, and the embryos were exposed to AgNWs for 72h. The development stage and mortality were measured every 24 hours for 72h. The developmental abnormalities measured were antennae, eye, rostrum, heart, carapace, post abdominal claw, malpighian tube, sensory bristles, and tail spine. The results show that the antennae and tail spin were the most sensitive to *D. galeata* embryonic organogenesis. The effect of effect on embryonic development with different lengths of AgNWs were negligible in the study.

MO291

Responses of antioxidant enzymes to nanoparticulate and ionic silver in aquatic microbes

D. Barros, Universidade do Minho / Centre of Molecular and Environmental Biology Department of Biology; A. Pradhan, University of Minho / Department of Biology; P.M. Santos, Universidade do Minho / Centre of Molecular and Environmental Biology CBMA Department of Biology; C. Pascoal, F. Cassio, University of Minho / Centre of Molecular and Environmental Biology (CBMA), Department of Biology

Silver nanoparticles (AgNPs) are among the most used nanoparticles and are likely to be released in significant amounts to aquatic environments. Due to their antimicrobial properties, it is relevant to examine whether AgNPs can pose a risk to aquatic microbes in natural ecosystems. Here, we investigated the effects of AgNPs (citrate-coated; 20 nm) on the growth and activity of antioxidant enzymes in i) two aquatic fungal strains of *Articulospora tetracladia*, one isolated from a non-polluted stream (AT72) and the other from a metal-polluted stream (AT61), and ii) the bacterial strain *Pseudomonas* sp. M1 (PsM1) isolated from a sediment sample in a metal-polluted stream. The activities of glutathione peroxidase (GPx), glutathione S-transferase (GST) and superoxide dismutase (SOD) were analyzed at AgNP concentrations inhibiting biomass production in 10% (EC_{10}) and 20% (EC_{20}) and the results were compared with those of ionic precursor (Ag^+ in $AgNO_3$). AT72 was the most sensitive strain to AgNPs, whereas PsM1 was the most tolerant one (EC_{20}: 7.5, 158.9 and 300 µg L^{-1} for AT72, AT61 and PsM1, respectively). The activity of all tested enzymes increased after exposure to AgNPs and Ag^+in all microorganisms. However, at similar stress level, the increase in all enzyme activities was higher in AT72 than in AT61 indicating higher oxidative stress in the strain from the non-polluted stream. The exposure to both stressors led to higher stimulation of GPx and SOD in PsM1, whereas SOD and GST responses were more pronounced in fungi. This suggests that the enzymatic defense mechanisms against both forms of silver might differ between bacteria and fungi.

\nFEDER-POFC-COMPETE and the Portuguese Foundation for Science and Technology supported this work (PIDDAC-PEst-OE/BIA/UI4050/2014, PTDC/AAC-AMB/121650/2010) and D. Barros (SFRH/BD/80407/2011).

MO292

Impacts of environmentally realistic silver nanoparticle exposure on Gammarus roeseli (Crustacea Amphipoda) from individual to functional levels.

J. Andreï, CNRS UMR 7360 / LIEC CNRS UMR; J. bouquerel, Universite de Lorraine; S. Pain-Devin, Université de Lorraine - UL / LIEC CNRS UMR; L. Giamberini, Université de Lorraine, CNRS UMR 7360 / LIEC CNRS UMR; F. Guerold, Universite de Lorraine / Laboratoire Interdisciplinaire des Environnements Continentaux

By means of some antibacterial properties, silver nanoparticles (nAg) are used in current consumer products and released into aquatic environment. One of the main priorities is now to address an appropriate risk assessment by using environmentally realistic exposure concentrations and media. Because they are good integrators of environmental perturbations, the gammarids are currently used as model organisms in ecotoxicology. They play a major role in ecosystem functioning by their capacity to breakdown leaf-litter and release fine particulate organic matter (FPOM) in the aquatic ecosystem. The objective of this study was to evaluate the impact of low concentrations of nAg in gammarids on both the locomotion behavior and the respiratory response. Ventilation of gills provides oxygen for cellular bioenergetics ensuring sufficient ATP levels to perform maintenance, reproduction and growth. On the opposite, the locomotion behavior is an ATP-dependent process that may be primarily impaired when energy lacks. Thus monitoring these parameters could provide integrative information about nanoparticles effects. In this context, the effects of low concentrations (0.5, 2.5 and 5µg.L^{-1}) of nAg of different sizes (10, 20, 40, 60 and 100nm in diameter) with the same shape and coating were assessed on the *G. roeseli* species after 36 and 72h of exposure. The locomotor activity was monitored in a group of 8 gammarids. After that, we measured individually and on the same organism the nAg impacts on: the oxygen consumption, the ventilatory activity and the electron transport system (ETS). Concurrently, measures at the functional level such as the organism's consumption and the FPOM production were performed. Realistic nAg concentrations did not induce mortality of gammarids despite a metal bioaccumulation. The first results showed an impact of the nAg on the gammarid oxygen consumption dependent of the nanoparticle size and concentration. The exposure to decreasing nAg sizes induced a proportional increase of the oxygen consumption by gammarids. For the 10nm nAg contamination condition, the highest concentrations induced also an increase of gammarid respiration. For environmentally realistic concentrations, nAg contamination has direct effects on organism's respiratory behavior. These impacts are dependent of the nanoparticle size and concentration. Some potential effects on the energy metabolism of *G. roeseli* could affect the aquatic ecosystem functioning.

MO293

The influence of pH and media composition on suspension stability of Ag, ZnO, and TiO2 nanoparticles and immobilization of Daphnia magna under guideline testing conditions.

D. Cupi, DTU (Technical University of Denmark) / Department of Environmental Engineering; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; A. Baun, DTU Environment / Department of Environmental Engineering

In aquatic toxicity testing of engineered nanoparticles (ENPs) the process of agglomeration is very important as it may alter the bioavailability of the ENPs and hence their toxicity. In this study we evaluated test conditions that are more favorable in maintaining a stable and low agglomerate size profile of ENPs in aquatic media applicable in OECD guideline. In this study we focus on controlling stability (as point of zero charge) by employing changes in pH to media of different ionic strength (M7, and Very Soft EPA medium) and documenting the influence of these parameters on acute immobilization of *Daphnia magna*. Despite being sterically stabilized, test suspensions of Ag NPs were found to consist of large agglomerate sizes (close to µm range) for both VS EPA and M7 media. The toxicity of the AgNPs was found to be higher in VS EPA medium than in M7 medium caused by an increased dissolution in VS EPA medium. Maintaining a constant pH throughout the testing period of 48 hrs proved to be challenging. Especially for ZnO NPs a constant and stable pH is crucial since a large degree of dissolution was observed below pH 6.5. Pzc for ZnO ENPs was observed at pH ~8. Increased dissolution of ZnO NPs tested in VS EPA medium (at pH 7) rendered them more toxic compared to M7 medium. TiO_2 ENPs revealed a pzc at pH values between 7-8 and were present in relatively low-size agglomerates (~200 nm) in VS EPA medium (pH 7), whereas the agglomerate size in M7 medium reached µm range. While various studies have classified TiO_2 ENPs non-toxic, we saw that the low-size stable agglomerates caused immobilization to *D. magna* revealing EC50